degummed silk whether degummed by conventional industrial degumming methods or by enzymatic degumming. Ammonia or ammonium ions were also effective when included as a component of the buffer used for enzymic degumming. Thus any of these methods of treatment of silk with ammonia or ammonium ions could be used to reduce the temperature, or the time, or the concentration of the chaotropic agent required to dissolve the silk resulting in reduced damage to the fibroin and a saving in process costs.

[0163] Treating *B. mori* silk with ammonia or ammonium ions enabled the time for dissolving the silk in 9.3 M lithium bromide solution at 60° C. to be cut from several hours to 15 minutes. Alternatively, ammonia or ammonium ion treatment enabled 7M lithium bromide to be used in place of 9.3 M at 60° C. It also enabled the silk to be completely dissolved in 9.3M lithium bromide solution at 20° C. within 24 hours. It further enabled the silk to be completely dissolved in 9.3M lithium bromide at 37° C. within 4 hours.

[0164] Therefore, it was found that treatment with ammonia or ammonium enables a range of milder treatments in which the temperature, concentration of the chaotropic agent or time required for solution can be varied singly or in combination. These milder treatments resulted in more rapid gelling times for the fibroin solution and stronger stiffer materials at the end of the process.

[0165] It is currently considered that another pair of ions with the same size, for example, potassium chloride will also have the same effect and could be used in place of the ammonia. This is supported by two lines of evidence: (1) The Jones-Dole viscosity (a measure of the chaotropicity) of potassium and chloride ions are similar as is the charge density enabling the ions to form ion pairs and help to remove an inner water shell of the protein (properties shared with ammonium chloride; and (2) Potassium chloride has been used to "salt in" proteins at salt concentrations generally ranging from 50 mM to 600 mM.

[0166] It is currently considered that certain other ionic reagents comprising an aqueous solution of monovalent cations and monovalent anions could provide the same effect. Particularly, it is thought that an ionic reagent comprising monovalent cations and monovalent anions having ionic radii of at least 1.3 Angstroms and a Jones-Dole B coefficient of between −0.05 and +0.1 at 25° C., would provide the same effect as the described in relation to the ammonium ions.

[0167] Suitable ionic reagents may include aqueous solutions of ammonium hydroxide, ammonium chloride, ammonium bromide, ammonium nitrate, potassium hydroxide, potassium chloride, potassium bromide and potassium nitrate.

[0168] Drying

[0169] The silk or silk cocoons are air dried overnight at room temperature in less than 20% humidity and in the presence of anhydrous calcium chloride.

[0170] The removal of substantially all of the water through drying increased the concentration of the ions in the solution, which was thought to enhance the effects of the ions and the resultant material.

[0171] Other known methods of drying such as freeze drying and drying through the application of heat would achieve the same effect. If heat drying is used, a temperature of less that 100° C. is thought to result in an improved fibroin material.

[0172] Degumming

[0173] The choice of the degumming method was also found to be crucial for the gelling time of the fibroin and stiffness and strength of the final material. Commercial reeling and degumming processes both use temperatures of around 100° C. and the use of sodium carbonate and/or Marseille's soap and it was found that reeled raw silks and degummed silks dissolved less readily than cocoon silks probably as a consequence of this treatment.

[0174] Degumming with commercial alcalase (bacterial subtilisin) enabled the degumming temperature to be reduced to 60° C. Alcalase is a member of the Serine S8 endoproteinase family and is likely to degrade fibroins badly as it has a broad specificity with a preference for a large uncharged residue in the P1 position. *B. mori* and *Antheraea pernyi* heavy chain fibroins have many predicted cleavage sites for this enzyme. The susceptibility of *B. mori* fibroin to alcalase cleavage was confirmed by polyacrylamide gel electrophoresis of a regenerated fibroin solution prepared from alcalase degummed silk.

[0175] In the case of degumming using trypsin the temperature for degumming could be reduced to 20-40° C. and gave gels with reduced gelling times, and with improved stiffness and strength compared with conventional high temperature degumming procedures. In contrast to alcalase, the tool PeptideCleaver showed few predicted trypsin cleavage sites in the consensus sequence of the repetitive crystalline domains and of the hydrophilic spacers of *B. mori* fibroin heavy chain fibroin and none in the consensus sequence or hydrophilic spacer in *A. pernyi* heavy chain fibroin. This suggested that it might be beneficial to degum silks in trypsin for the preparation of regenerated silk solutions. Trypsin was indeed found to be highly advantageous for degumming silk for the formation of improved regenerated silk solutions.

[0176] Silks degummed with trypsin gave regenerated silk solutions with shorter gelation times and capable of forming stiffer hydrogels than those obtained from regenerated silk prepared from silk degummed with alcalase. Degumming with trypsin gave gelling times of less than 5 minutes on exposure to glacial acetic acid vapour and also gave the stiffest and strongest materials suggesting that trypsin under these conditions produced much less chain cleavage than alcalase treatment.

[0177] It will be understood that other proteolytic enzymes producing little or no cleavage of fibroin may also be advantageous for degumming silks for the preparation of improved regenerated fibroin solutions. The observation that *B. mori* heavy chain fibroin contains very little proline while this amino acid is relatively abundant in sericin suggested that proline endopeptidase would be an ideal candidate to selectively remove sericin while producing little or no damage to fibroin.

[0178] Dialysis

[0179] In the course of these iterations it was found to be highly beneficial to dialyse regenerated fibroin solutions against type I milliQ™ water (available from Millipore™, 290 Concord Road, Billerica, Mass. 01821, US) otherwise known as ultrapure water, to remove the chaotropic agent from the silk solution.

[0180] It was noted that PIPES or Tris buffers or impurities in deionised water adversely affected the stiffness and strength of the final product when used as dialysants. It was noted that the inclusion of PIPES or Tris buffers or impurities in the dialysant also increased the viscosity of the regenerated